INCREASED SIZE AND NICOTINE PRODUCTION IN SELECTIONS FROM INTRASPECIFIC HYBRIDS OF NICOTIANA RUSTICA [1]

By HAROLD H. SMITH, *assistant geneticist,* and CHARLES W. BACON, *physiologist, Division of Tobacco Investigations, Bureau of Plant Industry, United States Department of Agriculture*

INTRODUCTION

At present nicotine is obtained from leaf stems and low-grade or scrap-leaf material of *Nicotiana tabacum* L. as a byproduct of the tobacco industry. However, with the possibility of an increasing demand for nicotine, because of its wider use in insecticides and for other purposes, there may be need for varieties of tobacco that can be grown profitably for the nicotine alone.

As early as 1911 *Nicotiana rustica* L. was shown [2] to have a higher content of nicotine than *N. tabacum*, and it was suggested at that time as a logical species to use as a source of this alkaloid.

For a number of years many different varieties and introduced strains of *Nicotiana rustica* have been grown by the Division of Tobacco Investigations and tested for yield and alkaloidal content. Types with the highest yields of nicotine were selected and continued each season. It was considered, however, that even among the best of these there was need for improvement of plant characteristics and especially for larger size.

The late Otto Olson, of this Division, working in cooperation with the Pennsylvania Agricultural Experiment Station, crossed two favorable varieties of *Nicotiana rustica* some years ago, and selections from his hybrids have also been continued.

With these sources of material the investigations reported herein were begun in 1936. Their purpose was to combine the best size characteristics to obtain a larger *Nicotiana rustica* giving increased yield of nicotine.

MATERIALS AND METHODS

Two inbred varieties from introduced seed lots and one of the best Olson selections were used as parents for crosses. They are described below.

(1) *Nicotiana rustica* var. *brasilia* Schrank, strain 34753 (fig. 1, *A*). Seed was obtained from A. Splendore, Scafati, Italy, in 1913, and was noted as coming originally from Brazil.

(2) *Nicotiana rustica* tall type (fig. 1, *B*). Seeds were obtained in 1926 from the Cambridge Botanical Garden, Cambridge, England.

[1] Received for publication March 3, 1941.

[2] GARRAD, G. H. THE GROWING OF TOBACCO FOR NICOTINE EXTRACTION. Southeast. Agr. Col. Jour. 20: 367-393. 1911.

They were not labeled with any varietal designation and have been called simply tall type. The strain is probably nearest to *N. rustica* var. *scabra* Cav.

(3) *Nicotiana rustica* Olson 68 (fig. 1, *C*). This line was selected from a cross between *N. rustica* var. Winnebago and *N. rustica* var. *brasilia* No. 23. It was continued by inbreeding through a number of generations until in 1938–39 the F_{10} and F_{11} were grown.

All plants in these experiments were grown at the Arlington Experiment Farm, Arlington, Va. They were placed 18 inches apart in 1-meter rows so that there were 8,800 to the acre. Shortly before planting time, 4–8–4 fertilizer was distributed uniformly in the drill

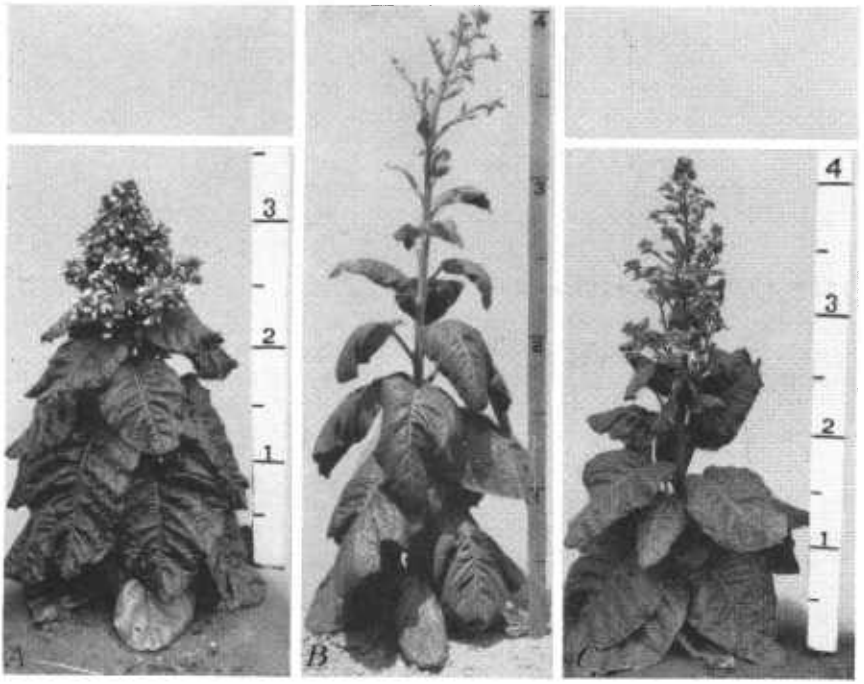

FIGURE 1.—Parental types of *Nicotiana rustica* used in crosses: *A*, Variety *brasilia*, strain 34753; *B*, tall type; *C*, Olson 68.

at the rate of 800 pounds per acre. The soil was Keyport silt loam, a heavy, claylike type, and was approximately uniform throughout the plots used.

Measurements on plant height, number of leaves, and length and width of the largest leaf were made soon after the onset of flowering. The best plants were bagged, and about 28 days later their leaves were picked and hung in the curing barn to dry.

Following the drying, or curing, selected leaf samples were weighed and ground into a fine powder preparatory to chemical analysis. A modification of the Keller method, developed in the Division of Tobacco Investigations, was used to determine the nicotine content.[3]

[3] GARNER, W. W., BACON, C. W., BOWLING, J. D., and BROWN, D. E. THE NITROGEN NUTRITION OF TOBACCO. U. S. Dept. Agr. Tech. Bul. 414, 78 pp., illus. 1934.

RESULTS ON INCREASING SIZE

PARENTS, F_1'S, AND F_2'S

Average measurements on the three parental types used are arranged in table 1. The year during which each family was grown is prefixed to its number; e. g., 37190 in 1937, 38179 in 1938. It is evident that pronounced yearly fluctuations occurred and must be taken into account when making comparisons between parents and hybrids.

TABLE 1.—*Means of measurements on parental strains of Nicotiana rustica and on F_1 and F_2 generation families resulting from crosses between them*

Parent or cross	Generation	Family No.	Number of individuals	Average plant height	Average number of leaves	Average length of largest leaf	Average width of largest leaf
				Inches		*Inches*	*Inches*
Variety *brasilia*, 34753	P_1	37190	38	28.5±0.31	19.5±0.31	13.2±0.21	10.4±0.23
	do	38179	20	29.9± .54	24.5± .82	13.1± .15	11.5± .14
	do	39232	5	14.4±1.78	11.8±2.35	9.7± .41	10.0± .65
Tall type	do	37192	68	38.4± .32	18.9± .29	12.2± .23	11.1± .29
	do	38200	24	50.0± .67	22.3± .33	12.9± .17	13.1± .29
	do	39233	5	46.0±1.30	20.2± .37	10.5± .45	10.9± .29
Olson 68	do	38186	33	32.1± .57	17.5± .52	12.9± .21	11.8± .13
	do	39238	5	30.0±2.55	14.0±1.95	10.9± .33	10.2± .41
34753 × tall type	F_1	37195	14	37.9±1.37	16.9± .27	12.6± .30	11.3± .34
	do	38203	5	47.4± .81	20.8± .58	14.3± .26	15.8± .20
(34753 × tall type) selfed	F_2	37196	70	39.2± .61	16.6± .35	13.3± .24	12.3± .19
(34753 × tall type) × 34753	do	37197	75	36.0± .86	17.7± .26	14.1± .24	12.7± .23
	do	38205	44	45.6±1.24	21.9± .16	14.6± .26	13.6± .29
(34753 × tall type) × tall type.	do	37198	71	40.5± .57	18.6± .32	12.2± .21	11.8± .22
(34753 × tall type) × Olson 68.	do	38222	80	51.9± .71	24.2± .47	14.9± .17	13.4± .16

The F_1 between strains 34753 and tall type was significantly taller than strain 34753, but was not as tall as the taller parent (table 1). It had significantly fewer leaves than either, and the largest leaf was at least as large as in the parents. Significance in this paper is determined by the t test for $P=0.05$. There was no decrease in average vigor from the first to the second hybrid generation. Backcrosses of the F_1 to strain 34753 (table 1) and to tall type tended to be intermediate between the F_1 and F_2 averages and those of the recurrent parent, with certain exceptions.

Some individuals with pronounced increases in size were obtained in the second hybrid generation. These were selfed and also were crossed with Olson 68 in an attempt to produce still larger plants. The immediate effect of Olson 68 on increasing plant height and number of leaves is evident by comparing (table 1) family 38205 with family 38222.

SELECTIONS FROM HYBRIDS

Four new lines were developed from the best plants in the second-generation families 37197 and 38222. They were continued by selfing and also were crossed once with Olson 68 and then successively selfed. Selections from the latter self-pollinations produced better plants than selections from the former. Average measurements on a few of the best families in each line, from the F_3 to the F_6, are shown in table 2. Data on some outstanding individuals are presented in table 3. The original F_2 plant of line 1 and its progeny were tall and had exceptionally large leaves, which was a major factor in producing the high yields of plant material obtained.

TABLE 2.—*Means of measurements on families in selected lines from hybrids between types of Nicotiana rustica*

Line No.	Generation	Family No.	Number of individuals	Average plant height	Average number of leaves	Average length of largest leaf	Average width of largest leaf
				Inches		*Inches*	*Inches*
Line 1	F_3	38208	69	48.2±0.98	21.2±0.45	15.5±0.19	14.2±0.20
	F_4	38224	72	51.2± .55	21.8± .25	16.3± .14	15.0± .15
	F_6	39293	5	52.2±2.58	25.2±1.59	13.1± .43	10.4± .43
Line 2	F_3	39245	8	46.3±3.03	23.8±1.54	11.9± .32	10.9± .52
	F_4	38228	42	44.6± .55	22.7± .58	15.1± .23	13.4± .20
	F_5	38278	19	50.4±1.52	27.5± .74	12.2± .22	9.6± .21
Line 3	F_4	38218	42	54.8± .83	28.0± .56	13.7± .13	12.0± .14
	F_4	38219	42	55.9± .64	27.9± .52	14.6± .18	12.4± .21
	F_6	39291	9	44.3±3.21	29.1± .83	11.3± .34	8.7± .28
Line 4	F_3	39271	10	41.7±1.27	22.9±1.07	11.7± .19	10.4± .43
	F_4	39292	6	58.7±1.48	25.0±1.16	11.7± .31	11.3± .34
Line 3 × line 1	$F_5 \times F_5$	39296	8	51.4±3.37	24.6± .87	12.9± .26	9.9± .26
Line 2 × line 1	$F_5 \times F_5$	39300	14	60.8±3.49	32.1±1.01	13.3± .26	10.7± .25

TABLE 3.—*Data on best plants in selections from hybrids*

Line and individual plant No.	Plant height	Number of leaves	Length of largest leaf	Width of largest leaf	Total weight of leaves	Nicotine, air-dry basis, plant not topped [1]	Estimated [2] potential yield of nicotine per acre if plants are topped
Line 1:	*Inches*		*Inches*	*Inches*	*Grams*	*Percent*	*Pounds*
38208–62	48	26	19.0	15.5	135.9	4.55	173
Line 2:							
39245–1	50	29	12.0	11.0	129.3	4.01	151
Line 3:							
38219–20	60	29	16.0	14.5	163.0	4.45	204
Line 4:							
38222–9	48	30	15.5	15.0	138.7	4.45	174
Line 3 × line 1:							
39296–1	36	23	12.5	9.0	112.4	5.30	159
Line 2 × line 1:							
39300–4	53	34	14.0	11.5	145.0	6.22	231
39300–6	61	41	13.5	11.0	129.0	4.90	173
39300–9	51	31	14.5	11.0	127.0	6.49	209

[1] Topping is the practice of cutting off the upper part of the plant, which includes the inflorescence and smallest leaves.

[2] Estimate based on a 2-percent increase in nicotine content if plants are topped. The yield from the stalks has not been added; this should amount to about 15 more pounds of nicotine per acre.

The original F_2 plant of line 2 and its progeny were large in all size characteristics measured and were late-flowering, a favorable feature. The original F_2 plant of line 3 and its progeny had relatively many leaves, which was a major factor in producing its high yields. Line 4 was developed from the best plant of family 38222, which was large and relatively high in nicotine content.

Many of the plants grown in 1939 were too stunted by the dry season to be considered typical; but when comparisons were made between families in which a limited number of representative individuals were measured, it was found that each new line was significantly larger than the best original types in one or more measurements. Increased size had been maintained, then, by selection and inbreeding through a number of generations and under adverse seasonal conditions.

In order to combine desirable characteristics of the new strains, crosses were made in all possible combinations among the best F_5 individuals in lines 1, 2, and 3 and the best F_5 in line 4. These F_1

crosses were each significantly larger (with one exception) than the original varieties. Especially promising was the cross between line 2 and line 1 (39300 in tables 2 and 3), which was superior to any of the new inbred lines, themselves. Line 3 × line 1 (39296) also proved to be a favorable combination.

In regard to the most outstanding segregants, plant No. 2 (fig. 2, *A*) of the cross line 2 × line 1 was 76 inches tall, which was the greatest height reached by any *Nicotiana rustica* grown during the course of

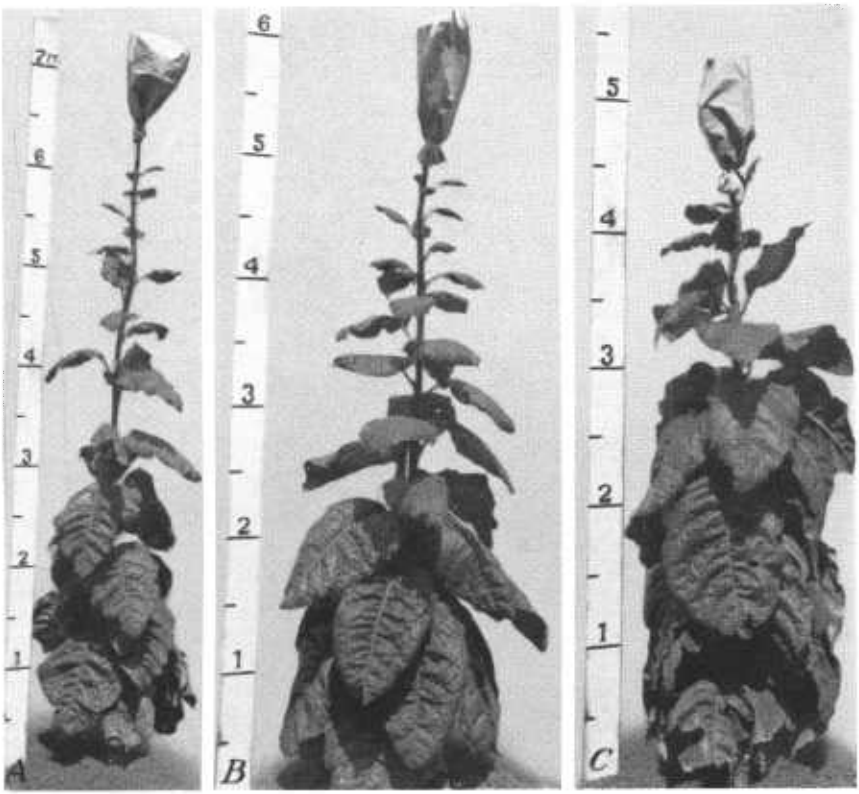

FIGURE 2.—Three exceptional plants in the family (39300) produced by crossing line 2 × line 1: *A*, Plant No. 2, height 76 inches; *B*, plant No. 6, 41 leaves; *C*, plant No. 4, 145 gm. of leaf material, estimated potential yield of 246 pounds of nicotine per acre.

the experiments. Plant No. 6 (fig. 2, *B*) of the same cross had 41 leaves, which was the highest leaf number recorded. Finally, the increased size of certain individuals was demonstrated by their high yield, which is discussed below.

RESULTS ON YIELD OF NICOTINE

WEIGHT OF LEAF MATERIAL

Selection in these experiments was based on yield of nicotine as well as on characteristics of the plant in the field. The former is mainly a function of total leaf weight and percentage of nicotine. Green plants

with the largest measurements in the field usually had the heaviest weight of dried leaf material, so that there was general correspondence between the two groups of data.

TABLE 4.—*Total weight of leaves, percentage of nicotine, and estimated potential yield of nicotine per acre from selected bagged plants of parental types of Nicotiana rustica*

Parental type	Family No.	Number of individuals	Total weight of leaves		Nicotine, air-dry basis		Estimated potential yield of nicotine per acre	
			Average	Highest	Average	Highest	Average	Highest
			Grams	*Grams*	*Percent*	*Percent*	*Pounds*	*Pounds*
Variety *brasilia*, 34753	37190	10	94.8	115.8	2.71	3.19	50.3	71.7
	38179	7	88.0	119.9	4.25	5.84	75.1	94.2
Variety *brasilia* No. 23	39231	4	41.2	63.9	6.15	7.24	47.9	68.4
Tall type	37192	9	74.5	91.2	2.09	2.73	31.5	48.3
	39233	4	44.2	50.5	3.44	4.11	28.7	34.1
Olson 68	38186	5	83.6	106.9	3.69	5.43	61.7	84.5
Olson 68A	39239	3	54.6	65.1	5.79	6.14	61.3	72.0

Weights for the parent strains are given in table 4, showing the average and maximum for each year. In 1939 data on strains 34753 and Olson 68 were not obtained, owing to an accident; so those on var. *brasilia* No. 23 and Olson 68A, two similar types, were substituted for comparison with hybrids grown that year.

TABLE 5.—*Total weight of leaves, percentage of nicotine, and estimated potential yield of nicotine per acre from F_1 and F_2 bagged plants of Nicotiana rustica hybrids*

Generation	Family No.[1]	Number of individuals	Total weight of leaves		Nicotine, air-dry basis		Estimated potential yield of nicotine per acre	
			Average	Highest	Average	Highest	Average	Highest
			Grams	*Grams*	*Percent*	*Percent*	*Pounds*	*Pounds*
F_1	37195	2	87.7	119.1	2.51	3.68	49.8	85.0
	38203	4	91.9	106.2	2.27	3.46	40.0	59.9
F_2	37196	12	98.0	120.8	3.19	4.20	61.5	86.3
	37197	24	103.2	132.2	3.11	4.22	63.2	108.2
	38205	10	95.7	125.2	3.54	4.99	66.5	104.3
	37198	16	96.8	143.6	2.85	4.56	55.6	127.0
	38222	15	101.2	143.5	3.65	5.57	72.0	119.7

[1] See table 1 for designation of parents for each family.

F_1's between strain 34753 and tall type (table 5) weighed about the same as strain 34753, the heavier parent. Best second-generation plants from the same cross were slightly heaver on an average than the F_1's and were outstanding in that the maximum weights were higher than in the parents or F_1's. This same result was obtained when germ plasm from Olson 68 was introduced (38222).

Plants in the new lines had the highest individual and average weights recorded during the experiments (tables 3 and 6). The dried leaf material alone, of the best plant in line 3, weighed 163 gm., which was the maximum weight obtained.

The best $F_5 \times F_5$ family, 39300, as judged by characteristics in the field, also had higher weights of leaf material than did any other

family grown that year. Leaves of the heaviest plant, No. 4 (fig. 2, *C*), weighed 145 gm., which compared favorably with the weight of the plants grown during better seasons.

TABLE 6.—*Total weight of leaves, percentage of nicotine, and estimated potential yield of nicotine per acre in bagged plants of best families in the new lines of Nicotiana rustica*

Line No.	Family No.	Number of individuals	Total weight of leaves		Nicotine, air-dry basis		Estimated potential yield of nicotine per acre	
			Average	Highest	Average	Highest	Average	Highest
			Grams	*Grams*	*Percent*	*Percent*	*Pounds*	*Pounds*
Line 1	38208	18	110.3	138.5	3.39	4.61	74.2	120.0
	39293	3	96.8	124.0	4.59	5.12	87.6	116.9
Line 2	39245	4	105.0	129.3	4.71	4.99	94.9	102.7
	39278	10	81.8	112.5	3.89	4.75	60.7	82.9
Line 3	38219	11	123.6	163.0	3.56	4.45	86.3	140.7
	39291	6	84.9	107.0	5.97	6.80	98.8	141.2
Line 4	39271	2	77.9	88.0	5.26	6.07	81.0	103.6
	39292	3	86.1	95.4	4.99	5.81	82.9	97.6
Line 3 × line 1	39296	4	95.3	112.4	4.10	5.30	76.9	115.6
Line 2 × line 1	39300	14	104.6	145.0	5.29	6.51	108.4	175.0

PERCENTAGE OF NICOTINE

Percentage of nicotine, like size of plant, is affected by the environment, so that pronounced yearly fluctuations occurred even in the homozygous parent strains.

Topping has a major influence on building up nicotine, and, from records over a 5-year period, it was found that topped plants had, on an average, an absolute increase of about 2.5 percent alkaloid as compared with bagged ones. An increase of 2 percent would be a conservative estimate of the effect of topping; this value corresponds closely to results obtained in New York State for 1924 and 1925.[4] Since all determinations in tables 4, 5, and 6 were made on bagged plants, approximately 2 percent should be added in order to estimate their potential content.

There was no marked gain or loss in percentage of nicotine from the higher parent to selected plants of the F_2 generation (table 5). Addition of Olson 68 in crosses caused an increase in nicotine production in both the F_2 and later generations.

The new lines were somewhat lower in percentage of nicotine than the parental types 34753 and Olson 68, but owing to their larger size some plants in most families were higher in yield of nicotine than the parents (table 6).

YIELD OF NICOTINE

Yield of nicotine per plant is the product of the weight and the percentage of nicotine. It was considered advisable to multiply this figure by the number of plants per acre, which would not change the relative individual values, and yet would provide some basis for estimating whether or not the increases were sufficiently great to be of possible economic importance.

[4] COLLISON, R. C., HARLAN, J. D., and STREETER, L. R. HIGH-NICOTINE TOBACCO. N. Y. State Agr. Expt. Sta. Bul. 562, 19 pp. 1929.

For bagged plants of the best parent strain during the years of the experiment an estimated maximum yield of about 94 pounds of nicotine per acre and an average of 75 were obtained (38179 in table 4).

The new lines contained bagged individuals with estimated yields as high as 141 pounds, and the average in a number of families was higher than in the parents (table 6). Fourteen selected bagged plants of the best $F_5 \times F_5$ family had an estimated maximum potential yield of 175 pounds of nicotine and an average of 108 pounds, obviously significant increases over the original varieties.

When the effect of topping and the yield from stalks are included, the potential values are still higher (see table 3, last column).

DISCUSSION

GENETIC CONSIDERATIONS

Increased height in intervarietal hybrids of *Nicotiana rustica* was reported by East [5] in crosses between the varieties *brasilia*, *texana*, *scabra*, and *humilis*. From the data obtained he concluded that reciprocal F_1's were very similar to each other and that the parent varieties differed by many genes for size. Hybrid vigor in first-generation plants from crosses between certain strains of *Nicotiana rustica*, but not others, was reported by Bolsunov [6] in Russia.

The F_1 described in these experiments was not larger than the parents, though other *Nicotiana rustica* crosses, less promising for nicotine production, did show typical hybrid vigor. The data reported present difficulties for genetic analysis, but certain general conclusions regarding the genetic basis for the results obtained appear tenable.

In crosses between strain 34753 and tall type it was found that the F_1 was almost as tall as the taller parent; the F_2 contained some plants that were taller than the parents or F_1; and in the F_4, families with significantly different mean heights were produced, one of which was significantly taller than tall type. These results, combined with observations on the backcrosses, were interpreted to mean that the parents differed by partially dominant genes for increasing height, most of which were from tall type. Some were contributed by strain 34753; so by genic recombination segregants and subsequent families were produced that exceeded the taller parent in height.

In crosses between strain 34753 and tall type it was found that the F_1 had fewer leaves than either parent. The F_2 generation did not differ significantly from the F_1 in average number of leaves and contained segregants that were equal, but no better, in leaf number than the original types. Backcrosses of the F_1 to each parent had significantly higher means for leaf number than the F_2. F_4 families with significantly different means occurred. Two F_4's and one F_6 had a significantly higher average number of leaves than the better parent.

It was concluded that increase in leaf number in these selections was controlled by genes, recessive in their action, some of which were contributed by each parent.

Tall type and strain 34753 do not differ significantly in length or width of the largest leaf. The F_1 was significantly larger according to 1938 results. The F_2 and first backcross generation showed a

[5] EAST, E. M. A STUDY OF PARTIAL STERILITY IN CERTAIN HYBRIDS. Genetics 6: [311]–365, illus. 1921.

[6] BOLSUNOV, I. I. CONTRIBUTION TO THE STUDY OF HYBRID VIGOUR IN NICOTIANA RUSTICA L. III DYNAMICS OF HYBRID GROWTH. Vsesoiuzn. Nauch. Issled. Inst. Tabach. i Makhor. Promysh. [Krasnodar] [Bul.] 139: [44]-57, illus. 1939. [In Russian. English summary, pp. 56-57.]

slightly increased variability over the F_1. Leaves of the backcross to strain 34753 were especially large. F_4 families were obtained that had significantly larger leaves than either parent. It was concluded that, although strain 34753 and tall type had approximately the same size largest leaf, the genes controlling this characteristic differed in the two types.

There was evidence that the genes controlling the inheritance of plant height, number of leaves, and size of the largest leaf were to some extent independent, an important factor in recombining desirable traits into new strains. The genotype of Olson 68 apparently contains more genes for increasing plant height than strain 34753; it can add to the number of leaves in certain genic combinations, and in no selections studied did the addition of Olson 68 cause a significant difference in leaf size.

In table 7, the frequency distributions of measurements on plant height, number of leaves, and length of the largest leaf are arranged for 34753, tall type, F_1, $F_1 \times 34753$, best F_4, and best $F_5 \times F_5$. The families involve only strain 34753 and tall type, with the exception of the best $F_5 \times F_5$, which contains germ plasm from Olson 68 as well as from the other two parents. Table 7 shows the variability in the families concerned and gives some idea of the effect of hybridization and of selection on the characteristics measured.

TABLE 7.—*Frequency distributions of plant height, number of leaves, and length of largest leaf for two parent types and selected hybrid families*

PLANT HEIGHT

Parent or hybrid	Frequency distribution of plant height in inches																									Mean height
	24	26	28	30	32	34	36	38	40	42	44	46	48	50	52	54	56	58	60	62	64	66	68	70	72	74	76	
34753	4	2	6	8	7	3																						29.9
Tall type										1	1	4	3	5	8	1	1											50.0
F_1												1	1	3														47.4
$F_1 \times 34753$				3	1	2	1	3	3	7	3	5	2	4	0	3	4	2	0	1								45.6
Best F_4									1	1	0	2	4	4	3	3	6	9	8	1								54.8
Best $F_5 \times F_5$				1	0	0	0	0	0	1	0	0	0	1	1	0	0	0	3	1	0	0	2	0	3	0	1	60.8

NUMBER OF LEAVES

Parent or hybrid	Frequency distribution of number of leaves															Mean number of leaves
	12	14	16	18	20	22	24	26	28	30	32	34	36	38	40	
34753	1	2	0	0	2	5	6	6	6	2						24.5
Tall type				1	6	10	7									22.3
F_1				1	2	2										20.8
$F_1 \times 34753$		1	6	2	10	8	12	4	1							21.9
Best F_4			1	1	1	0	4	10	13	5	6	1				28.0
Best $F_5 \times F_5$								1	3	3	1	5	0	0	1	32.1

LENGTH OF LARGEST LEAF

Parent or hybrid	Frequency distribution of leaf length in inches											Mean length
	10	11	12	13	14	15	16	17	18	19	20	
34753		2	5	15	8							13.1
Tall type		3	7	11	3							12.9
F_1				1	3	1						14.3
$F_1 \times 34753$	2	2	2	4	9	14	9	1	1			14.6
Best F_4	1	1	2	2	7	9	12	3	4	0	1	15.3

There were insufficient data to warrant drawing any conclusions regarding the genetic factors controlling nicotine content. It was evident, however, that either some genes for nicotine production from strain 34753 and Olson 68 were not included in the genotypes selected for the new lines, or else larger plants are not capable of building up as high a percentage of nicotine as smaller ones under the same conditions.

ECONOMIC CONSIDERATIONS

On the basis of prices paid for tobacco sold for byproduct, the returns per acre for crops containing 100 to 150 pounds of nicotine would compare favorably with the average returns over a period of years from culture of at least some types of commercial leaf tobacco. Topped plants of the original varieties actually gave such yields under the conditions described here, and the production of alkaloid may be increased somewhat by allowing the plants to remain longer in the field from time of topping to harvest. The costs of culture and of nicotine extraction are not definitely known, however, and it has been estimated that the profit, if any, from growing the original varieties for nicotine would be small.

The results on best selections in the new lines, to the F_6 generation, indicate that it may be possible to grow types of *Nicotiana rustica* that yield up to 200 pounds of nicotine per acre under the cultural and environmental conditions of these experiments. If the homozygous strains now being developed are able to produce as high yields in field tests, it should be possible to grow *N. rustica* profitably for the nicotine alone.

The cost of preparing *Nicotiana rustica* for market would be less than for commercial leaf tobacco; but, since it forms suckers more abundantly than *N. tabacum*, more labor would be required for their removal. In this connection, Bolsunov[7] has recently described an unusual variant, resulting from a cross between two types of *N. rustica*, in which the axillary buds were either partially atrophied or completely absent and in some cases of which the terminal inflorescence was also lacking. Seed was obtained from secondary suckers on the lower part of the plants, and selection to the F_5 generation has produced lines which still show the anomaly. This type, if it is as promising as described, should be of value, since the cost of topping and suckering would be reduced.

SUMMARY

New lines of *Nicotiana rustica*, developed by hybridization and selection, were significantly larger on an average than their three parental strains, namely, *N. rustica* var. *brasilia* (strain 34753), tall type, and Olson 68.

The characteristics measured were plant height, number of leaves, and size of largest leaf. These were controlled by relatively independent genes that could be recombined in advantageous groupings. The best parent values were usually not exceeded until new homozygous combinations were established in the F_2 or later generations. The original strains differed by some genes for each character.

[7] BOLSUNOV, I. I. [A VALUABLE HYBRID OF NICOTIANA RUSTICA DEVOID OF INFLORESCENCE AND UPPER SUCKERS.] Selek. i Semen. 2-3: 40-41, illus. 1939. [In Russian.]

Further increases in size were obtained by crossing F_5 selections of the new lines together. The occurrence of some exceptionally large segregants suggests that still further increases in the average size will be possible.

Related data on the weight of dried leaf material and percentage of nicotine were reported. Estimated potential yields of nicotine per acre were calculated from these figures, and selections in the new strains, to the F_6 generation, were found to be superior to their original parents in this respect. The possible economic importance of the improved lines, for use as a source of nicotine for insecticidal purposes, was discussed.